REGULATION OF NORMAL AND NEOPLASTIC HUMAN MAST CELL DEVELOPMENT IN MASTOCYTOSIS

DEAN D. METCALFE, M.D.

BETHESDA, MARYLAND

ABSTRACT

Mastocytosis, a condition characterized by a pathologic accumulation of clonal mast cells in tissues, offers a unique opportunity to study the growth and differentiation of mast cells as well as their contribution to various pathologic processes. This is because molecular pathways governing the proliferation and survival of mast cells show striking similarities between normal mast cells and their counterparts in mastocytosis. For example, activation of Kit, a transmembrane receptor for stem cell factor (SCF) is critical for the growth and differentiation of normal mast cells. Mutations such as D816V resulting in its pathologic activation are strongly associated with mastocytosis. Treatment of mastocytosis is aimed at controlling symptoms, as no specific drug has yet been clinically demonstrated to selectively eliminate mast cells carrying the D816V gain-of-function c-kit mutation. Non-myeloablative bone marrow transplantation is performed in select patients to take advantage of the immunotherapeutic effects of the graft.

Mast Cells and Systemic Mastocytosis

Mastocytosis has been perplexing clinicians for over a century. In 1869 Nettleship and Tay described in a 2-year-old girl what is now considered to be the most common skin lesion in mastocytosis, urticaria pigmentosa (UP) (1). In 1877, Paul Ehrlich was the first to describe mast cells in his study of connective tissue cells (2). He believed that these cells represented over nourished or overfed connective tissue cells and termed them mastzellen. Near the same time, Unna demonstrated mast cells in the skin lesions of UP (3), the term suggested by Sangster the following year (4). In 1949 the systemic nature of mastocytosis was recognized by Ellis in his report of the autopsy findings of a 1-year old infant with diffuse organ infiltration by mast cells (5).

While the most commonly involved organ in systemic mastocytosis is

Address: NIH/NIAID/LAD, Building 10, Room 11C205, 10 Center Drive, MSC 1881, Bethesda, MD 20892-1881, Phone: 301-496-2165, Fax: 301-480-8384, E-mail: dmetcalfe@niaid.nih.gov.

the skin, a significant number of cases eventually show mast cell infiltration in multiple organs. It is in this clinical setting that the term systemic mastocytosis has generally been used. This systemic organ infiltration by mast cells often results in protean clinical manifestations that frequently obscure the underlying disease process. A consensus classification schema adopted by the World Health Organization in 2001 (6) is widely employed as a reasonable approach to the classification and therefore the diagnosis of this disease (Table 1).

Biology and Pathogenesis of Mast Cell Diseases

Mast cells are considered the initial tissue-based effector cells of the immediate allergic reaction by virtue of their high-affinity receptors for IgE. The wide distribution of mast cells throughout the human body, especially in proximity to blood vessels in atopic and nonatopic individuals alike, and the recognition of mast cell activation by non-IgE and nonimmunologic means with release of biologically active mediators such as histamine, leukotrienes, and cytokines, suggests that these cells also may be involved in nonatopic diseases and in homeostasis. Thus, mast cell-mediated events have been postulated to play a

TABLE 1
*WHO—Classification of Mastocytosis**

Variant	Subvariants
Cutaneous Mastocytosis (CM)	
	—Urticaria Pigmentosa (UP) = Maculopapular CM (MPCM)
	—Diffuse CM (DCM)
	—Mastocytoma of Skin
Indolent Systemic Mastocytosis (ISM)	
	—Smouldering SM
	—Isolated bone marrow mastocytosis
Systemic Mastocytosis with an associated clonal haematologic non mast cell lineage disease (SM-AHNMD)	—SMAML
	—SM-MDS
	—SM-MPD
	—SM-HES
	—SM-CMML
	—SM-NHL
Aggressive Systemic Mastocytosis (ASM)	
	—Lymphadenopathic SM with eosinophilia
Mast Cell Leukaemia (MCL)	
	—Aleukaemic MCL
Mast Cell Sarcoma	
Extracutaneous Mastocytoma	

* For details of the WHO classification of mastocytosis, see ref 6.

role in innate and acquired immunity, in wound healing and in tumor angiogenesis.

It is now known that mast cells are derived from pluripotential hematopoietic cells (31). This was first demonstrated with in vivo experiments using genetically mast cell-deficient mutant mice and their co-geneic normal littermates (7,8). Human mast cells may now be cultured from CD34+ cells from blood or bone marrow in sufficient numbers to permit the study of mast cell growth and differentiation (9). As in rodents, Kit and its ligand SCF are involved in the growth and differentiation of human mast cells. In the human, SCF is encoded by a gene on chromosome 12. SCF is primarily, but not exclusively, produced by stromal cells (10). SCF may either be released as a soluble growth factor, or it may be expressed on the cell surface of these cells. Other cytokines in the presence of SCF such as IL-6 may further increase cell numbers.

While mast cell precursors originate in the bone marrow, they migrate to tissue sites and mature. Under normal conditions, mast cells are found throughout vascularized tissues and are particularly numerous associated with the epithelial surface of the skin and respiratory tract, in the gastrointestinal and genitourinary tracts, and in perivascular areas (11). At these sites, mast cells exhibit one of two predominant phenotypes, as defined by staining characteristics and granule enzyme content. Most mast cells within the skin and other connective tissue sites stain intensively with dyes, probably due to their heparin content. Historically, these mast cells have been referred to as "connective tissue" mast cells. Mast cells at mucosal locations, such as the lamina propria of gastrointestinal tract, stain less intensively and are referred to as "mucosal" mast cells. In humans, the mucosal phenotype contains a specific tryptase, while the connective tissue phenotype contains both this tryptase and a chymotryptase (12). The human mucosal mast cell is sometimes referred to as the "T mast cell" for its tryptase content, and the human connective tissue mast cell as a "TC mast cell" for its content of both tryptase and chymotryptase.

It is now accepted that activating mutations in *c-kit* are associated with an increase in mast cell numbers in most adult patients with mastocytosis, and in a subset of children with more severe disease. The original observation was suggested by the report that CD34+ cells from the peripheral blood of patients with mastocytosis, when cultured in vitro in the presence of SCF, gave rise to higher numbers of mast cells per CD34+ cell compared with cells from normal subjects (13). A search for activating mutations in *c-kit* based on this observation led to the identification of the somatic mutation Asp816 to Val in *c-kit* in

peripheral blood mononuclear cells (PBMC) from patients with mastocytosis (14). This mutation was subsequently identified in the skin and tissues of mastocytosis patients. More recently, additional activating mutation in *c-kit* at codon 816 have been identified including Asp 816Phe and Asp816Tyr (15). The Asp816 Val mutation has been detected in T cells, B cells, and myelomonocytic cells in patients with mastocytosis, consistent with the conclusion that mastocytosis is a clonal hematopoietic stem cell disorder (16).

It has also been suggested that, apart from D816V, other *c-kit* mutations can contribute to the development of MC disorders (17–19). Notably, in patients with SM, several c-kit mutations have been identified (Table 2) (20). In addition, various chromosomal defects, other gene defects, and genetic polymorphisms have been discussed as contributing to the pathogenesis of SM (21–24). Many of these defects are detected in patients who have an additional myeloid neoplasm apart from SM, i.e. an associated haematopoietic clonal non-MC lineage disease (AHNMD). Most of these defects have recently been linked to distinct myeloid neoplasms and have been defined as disease criteria by the WHO. Other defects, such as the FIPL1/PDGFRA fusion gene, have been associated with specific myeloid neoplasms and judged as reliable markers sufficient to define a certain subtype of AHNMD in

TABLE 2
Gene Defects, Gene Polymorphisms, and Karyotype Abnormalities Reported Associated with Mastocytosis

Finding	Disease Association	% (estimated)
Gene defects		
c-kit D816V	all variants of SM (and in some with CM)	>80%
c-kit D816Y	CM, SM, SM-AHNMD	<5%
c-kit D816F	CM	<5%
c-kit D816H	SM-AHNMD	<5%
c-kit D820G	ASM	<5%
c-kit V560G	SM	<5%
c-kit F522C	SM	<5%
c-kit E839K	CM	<5%
c-kit V530I	SM-AML	<5%
c-kit K509I	SM (familial type)	<5%
FIPL1/PDGFRA*	SM-HES, SM with eosinophilia	<5%
Gene polymorphisms		
IL-4Rα Q576R	CM, indolent SM (ISM)	n.k.
Karyotype abnormalities		
del 20(q12)*	SM, SM-AHNMD	<5%
+9	SM, SM-AHNMD	<5%
t(8;21)*	SM-AML M2	<5%

* These gene abnormalities are indicative of an AHNMD.

patients with SM. Thus, the presence of eosinophilia and FIPL1/PDGFRA in a patient with SM aids in the diagnosis of SM with hypereosinophilic syndrome (SM-HES), a special variant of SM-AHNMD.

A second important pathogenetic aspect in SM is the abnormal expression of cell surface adhesion antigens on neoplastic MCs (25). Several antigens specifically detectable on neoplastic mast cells in SM, such as CD2 (LFA-2), represent cell-cell adhesion molecules. Whether these molecules, especially CD2, indeed play a pathogenetic role in SM remains unknown.

Pathobiology

The pathobiology of mast cell disease can be divided into systemic and local disease. The systemic effects of this disorder arise from the release of significant amounts of mediators into the systemic circulation which may result in such findings as flushing or syncope. The local manifestations of this disease result largely from the effects of local collections of mast cells. Other systemic effects of mast cell disease result largely from severe end-organ dysfunction (e.g. hepatosplenomegally, bone marrow dysfunction).

The classic pathologic lesion of cutaneous mast cell disease is the lesion of UP. However, in at least one-third of cases of systemic mastocytosis, cutaneous lesions are lacking, and other organs need to be biopsied to make the diagnosis. The most frequently involved organs in systemic mastocytosis are the bone marrow, skin, lymph nodes, spleen, liver, and gastrointestinal tract; the lungs and kidneys are virtually never involved. Because of the high frequency of bone marrow involvement and the relatively high frequency of bone marrow biopsy and aspirate in the diagnosis of this disorder, this procedure has become the method of choice in establishing systemic disease.

Focal mast cell lesions of the bone marrow have been reported in up to 90% of adults with systemic mast cell disease (26). The typical bone marrow lesions consist of foci of spindle-shaped mast cells in a fibrotic background. Usually there is an abundant admixture of mast cells and T- and B-lymphocytes, sometimes associated with eosinophils. These lesions may be located in perivascular, peritrabecular, and intertrabecular sites. In marrows extensively involved by mast cell lesions, the bony trabeculae are often moderately to markedly thicken.

While some patients with systemic mast cell disease may have a marked increase in marrow mast cells in the absence of focal mast cell lesions (27), most patient marrows contain focal mast cell lesions. The procedure of choice to visualize mast cells in marrow sections is by using antibody to human mast cell tryptase (6). The typical mast cell in

the biopsy sample has a spindle-shaped or oval nucleus and prominent granules. Mast cells in aspirate smears are elongated or spindle-shaped. Normal mast cells in aspirate smears appear round. In comparison to adults with systemic mast cell disease, marrow involvement in children appears to be much less common (28).

The progression of marrow involvement in systemic mast cell disease is unknown. While some adults appear to have stable, or possibly decreasing, marrow involvement over time, the clinical significance of the extent of marrow involvement by mast cells remains elusive. What is known, however, is that adult patients may experience resolution of lesions of UP even while bone marrow involvement progresses (29).

While lymph nodes, liver, and spleen are infrequently biopsied in patients with systemic mast cell disease, pathologic studies have documented significant involvement in these organs. Lawrence and colleagues found lymphadenopathy in 25% of patients with systemic disease at presentation but in none of the patients in whom the disease was confined to the skin (30). In a study of 21 patients, Horny et al. reported lymph node involvement in 80% with the medullary cords and sinuses most often involved (31). In reviewing the records of 58 mastocytosis patients followed at the Mayo Clinic, Travis and Li demonstrated peripheral lymphadenopathy in 26% and central lymphadenopathy in 19% of patients at diagnosis (32). Lymphadenopathy was more pronounced in patients with associated hematologic malignancies and aggressive nonleukemic mastocytosis. Within the lymph nodes, mast cell infiltrates were most common in the paracortex, followed by the follicles, the medullary cords, and the sinuses.

Spleen involvement is also common in mastocytosis, seen in 40–50% of patients with systemic disease at presentation (30). Travis et al. reviewed the pathologic features (32). A paratrabecular distribution of mast cell infiltrates was most common. Also observed were perifollicular, follicular, and diffuse patterns. Trabecular and capsular fibrosis and eosinophilic infiltration were present in the biopsies examined along with extramedullary hematopoiesis.

Liver disease with mast cell infiltration is a common finding in patients with mastocytosis. Severe liver disease is uncommon, except in patients within aggressive categories (those with an associated hematologic disorder or aggressive mastocytosis) (33). Elevated serum alkaline phosphatase, serum aminotransamidases, 5′ nucleotidase, or gamma-glutamyltranspeptidase (GGTP) was often detected. Patients with aggressive disease were more likely to develop ascites or portal hypertension and to die of complications of mastocytosis. In these

patients, portal fibrosis and venopathy and veno-occlusive disease was usually observed.

Clinical Manifestations

The WHO classification for systemic mast cell disease is clinically useful in categorizing the variants of this disease (Table 1) (6). This schema divides systemic mast cell disease into five categories of increasing clinical aggressiveness as well as extra cutaneous mastocytoma. The first systemic category is termed "indolent" systemic mastocytosis (ISM). Most mastocytosis patients with systemic disease seen will fall into this category. Their disease eventually involves the liver, spleen, lymph nodes, GI tract, and bone marrow. The pathophysiologic process may be managed successfully for decades and does not appear to shorten life span. Manifestations of ISM include: hemodynamic instability, cutaneous mast cell disease, ulcers of the stomach and duodenum associated with increased gastric acid, malabsorption due to systemic mediators and mast cell infiltration of the intestine, mast cell infiltration of the bone marrow, skeletal disease due to the activity of mast cells on bony surfaces, hepatosplenomegaly, and lymphadenopathy due to mast cell infiltration. These manifestations do not define subgroups, since a particular patient may have more than one manifestation.

The second category of systemic disease consists of systemic mastocytosis with associated clonal non-mast-cell lineage disease (SM-AHNMD). Affected patients have increased mast cells in one or more target organs plus a demonstrable bone marrow abnormality such as a myeloproliferative or myelodysplastic disorder; the skin is variably involved. In this category, the prognosis is determined primarily by the associated hematologic disorder. The third category, aggressive systemic mastocytosis (ASM) typically is a rapidly progressive disease involving first the bone marrow and then the gastrointestinal tract, liver, spleen, and lymph nodes. The prognosis is much more guarded than that of the first category. The next category of systemic disease is mast cell leukemia (MCL), a primary leukemic process with increased mast cell burdens in both bone marrow and blood. The prognosis for patients with MCL is extremely poor and survivals are typically <1 a year, even with aggressive combination chemotherapy (34). Mast cell sarcoma (MSC) is exceedingly rare and is characterized by a destructive growth of a tumor made up of highly atypical immature mast cells. An extracutaneous mastocytoma is very rare, and usually localized to the lung (6).

The typical clinical presentation for patients with a systemic mast cell disorder is hard to define, as any patient may have more than one organ involved with mast cells. In addition, many patients exhibit

vague or nonspecific constitutional symptoms such as weakness, fatigue, night sweats, and weight loss that cannot be attributed to any particular organ dysfunction. Some patients present with neurologic or neuropsychiatric manifestations, including alterations in cognitive abilities, or depression. The etiology of such symptoms is unclear, although mediator-induced hypotensive effects on the brain or a mixed organic brain syndrome, or both, have been hypothesized (35).

Some patients describe "attacks" characterized by flushing or sensations of warmth, usually accompanied by palpitations, shortness of breath, chest discomfort, nausea and diarrhea, headache, lightheadedness, and occasionally hypotension. Syncopal episodes may be precipitated by heat exposure, emotional stress, or physical exertion. During episodes of flushing, many patients experience a significant decrease of blood pressure accompanied by tachycardia.

Cutaneous Disease

Cutaneous manifestations of disease may appear early in life (26,29,36,37). The onset of UP follows a biphasic curve with one peak at 2.5 months of age and the second peak at 26.5 years (36). Lesions are present in 80% of affected children by 6 months of age. Skin lesions are reported in 50–100% of patients with systemic mastocytosis. A familial association is rare. In approximately 50 families, mastocytosis of one form or another has been reported to affect more than one family member, including several families with affected twins (38–41). Mast cell disease has four cutaneous manifestations: UP with variants including telangiectasia macularis eruptiva perstans (TMEP), diffuse cutaneous mastocytosis (DCM) and mastocytoma. The most common cutaneous mast cell lesion is typical UP, which appears as red-brown macules, papules, and plaques. Lesions occur in a generalized and random distribution. Erythema, edema, and blister formation with subsequent crusting of the lesions has been reported, particularly in young children. After the age of 10, vesicles generally do not occur, and the lesions tend to be smaller and more numerous. Approximately one-half of patients in whom UP appears in infancy or childhood experience resolution by adolescence; in the remainder of patients, only lightly pigmented macules remain (36,42,43). Lesions that appear after age 10 tend to persist and remain symptomatic. Telangiectasias, petechiae, or ecchymosis may occur in the lesions or in adjacent clinically normal skin. The most common clinical manifestations include pruritis, dermatographism, and the presence of a Darier sign (wheal and erythema occurring after a brisk stroke to a lesion).

Solitary mastocytomas of skin may be present at birth, although most

appear within the first 3 months of life and are rarely described in adults (42). Most mastocytomas form on the extremities and rarely involve palms or soles. In most cases, the lesions are thought to involute spontaneously; however, this has not invariably been reported. Rare patients have had systemic manifestations such as flushing.

Diffuse cutaneous mastocytosis (DCM) is a rare disorder that generally presents before the age of 3 and involves the entire cutaneous integument. The skin may appear normal; more commonly, it has a yellow-red-brown color with a peau d'orange appearance (42). Dermatographism with the formation of hemorrhagic blisters is a common finding. DCM reportedly may resolve spontaneously at age 5–15 months; in other cases the disease persists. Children with DCM are at risk of complications such as flushing, hypotension, gastrointestinal bleeding, shock, and occasionally death.

Telangestasia macularis eruptive perstans (TMEP) is a rare form of mastocytosis traditionally thought to be limited to the skin (42). The lesions in this disorder are generalized, red, telangiectatic macules on a tan to brown background. Individual lesions are 2–6 mm in diameter and are without sharply defined outlines. Sites become edematous when rubbed. In occasional patients, the lesions may coexist with those of UP.

Hematologic Abnormalities

In 90% of patients with systemic mastocytosis, either a focal or diffuse increase of mast cells in the bone marrow will be identified. A number of hematologic abnormalities are reported in patients with systemic mast cell disease, although one or more of the abnormalities is not always present. Anemia is the most common finding, occurring in one-third to one-half of all patients. Thrombocytopenia and leukopenia have been demonstrated in roughly 15–20% of patients. Leukocytosis has been demonstrated in 20–30% of patients, and eosinophilia in up to 40% of patients with systemic mast cell disease. Monocytosis has been reported in approximately 15% of patients with mastocytosis. Lymphocytosis and thrombocytosis are unusual findings.

In more severe forms of disease, mast cells may be found in peripheral blood where they frequently have the appearance of atypical monocytes containing scattered large basophilic granules. They also frequently appear somewhat dysplastic, making identification on Wright stain difficult. Frequently, stains specific for mast cells must be performed to identify the cells in the peripheral blood conclusively and to distinguish them from either basophils or hypergranulated monocytes.

Patients with systemic mastocytosis may present with a hematologic syndrome resembling a myeloproliferative or myelodysplastic process. Several premalignant or overtly malignant syndromes have been described with systemic mast cell disease including the above disorders as well as lymphoma, acute myeloid leukemia, or a lymphoproliferative disease. The peripheral blood picture in these patients may be consistent with that of either chronic myeloid leukemia (CML) or chronic myelomonocytic leukemia (CMML). The presence of either of these disorders is associated with a poor prognosis (27,30). As with primary myeloproliferative and myelodysplastic syndromes, a secondary acute leukemia may develop. Many of the patients with systemic mast cell disease who are demonstrated to have significant cytopenias on bone marrow examination have overtly dysplastic myeloid or erythroid maturation. In addition, a small number of patients have developed diffuse fibrosis of the bone marrow with a marked hypocellularity. Those patients with systemic mast cell disease with a CML-like picture are typically Philadelphia chromosome negative.

Gastrointestinal Disease

Gastrointestinal symptoms have commonly been reported including abdominal pain, diarrhea, nausea, vomiting, and peptic ulcer disease in one-fourth to one-half of individuals with systemic mastocytosis. Cherner et al. demonstrated a broad range of basal acid secretion values in patients with systemic mastocytosis (44). Six of 16 patients were shown to have clinically significant acid hypersecretion. Up to one-third of patients have some evidence of fat malabsorption, although the degree of malabsorption is generally not of clinical importance. While the etiology of this mild malabsorption is unclear, potential explanations include acid hypersecretion and mucosal dysfunction. Shortened intestinal transit time does not appear to contribute.

Hepatic Disease

Hepatic involvement with mast cell lesions is a common finding in systemic mastocytosis, although a significant degree of hepatic dysfunction is generally not observed. Alkaline phosphatase is frequently elevated, but this elevation is largely attributable to bone disease. Webb et al. demonstrated hepatomegaly in 45% and splenomegaly in 50% of 26 patients they studied (26). Rare patients with systemic mastocytosis have been demonstrated to have portal hypertension (33), and significant splenomegaly may be a contributing factor to the hematologic abnormalities noted in many patients.

Lymphadenopathy

Lymphadenopathy has been demonstrated in up to 60% of patients with systemic mast cell disease (32). Both peripheral and central lymphadenopathy has been documented (30). The presence of lymphadenopathy in and of itself does not signal aggressive disease, and no specific symptoms are referable to lymphadenopathy. Patients who present with lymphadenopathy and significant hepatosplenomegaly should be followed closely for evolution into a more aggressive systemic disorder.

Skeletal Disease

The most common radiographic abnormality in patients with systemic mastocytosis is diffuse osteopenia (45). Both lytic and sclerotic lesions have also been described. Bone scans may be normal or may show focal or diffuse abnormalities (46). In patients with aggressive disease, diffuse musculoskeletal pain, as well as pathologic fractures have been reported.

Aggressive Systemic Mastocytosis

A subset of patients with systemic mastocytosis presents with, or subsequently develops, significant organ-function impairment due to infiltration by neoplastic mast cells. Associated findings include lymphadenopathy, hepatosplenomegaly, and peripheral eosinophilia (27,47). Lymph node biopsies in these patients frequently show a hyperplastic picture suggestive of a malignant lymphoproliferative disorder. However, tissue pathology does not support a diagnosis of lymphoma. Clinically, these patients have an aggressive form of the disease with marked visceral and bony involvement and dramatically shortened survival in the absence of development of an associated hematologic disorder.

Mast Cell Leukemia

These cases are characterized by the presence of a large number of atypical-appearing mast cells in the peripheral blood, a leukocytosis and granulocytosis, and a compressed clinical course (34). In most cases, multiorgan failure including bone marrow failure develops over weeks to months. The bone marrow shows a diffuse and dense infiltration of mast cell that replaces normal marrow.

Laboratory Evaluation and Diagnosis

If mastocytosis is suspected on clinical grounds, the routine workup should consist of examination of the skin, both gross and microscopic;

a bone marrow biopsy and aspirate; and serum for tryptase levels. Bone marrow aspirate mast cells from patients with mastocytosis often exhibit an aberrant immunophenotype in that they may express CD2 and CD25 in addition to CD117. Flow cytometry may thus be useful in identifying patients with this disease (6). Additional studies, as suggested by symptomatology or findings during the routine evaluation, may include a bone scan (46); a gastrointestinal evaluation involving radiographic studies of the upper gastrointestinal tract and small intestines, computed tomography scan, and endoscopy; and a neuropsychiatric workup.

The diagnostic criteria for mastocytosis include one major and four minor criteria; where one major and one minor or three minor criteria are required for diagnoses (Table 3). The classic lesions of mastocytosis consist of multifocal dense mast cell aggregates representing the sole major criteria. Minor criteria include atypical mast cell morphology, detection of codon 816 mutations in Kit, expression of CD2 and/or CD25 by bone marrow mast cells, and a total serum tryptase of >20 ng/ml.

While it is clear that symptoms of mast cell disease may reflect local increases of mast cells, slight increases in mast cell numbers in target

TABLE 3
Diagnostic Criteria for Mastocytosis[1]

Cutaneous Mastocytosis
Typical clinical findings of UP/MPCM, DCM or solitary mastocytoma, and typical infiltrates of mast cells in a multi-focal or diffuse pattern on skin biopsy.
Systemic Mastocytosis
Major criteria
Multifocal, dense infiltrates of mast cells (15 or more mast Cells in an aggregate) detected in sections of bone marrow and/or other extracutaneous organ(s), and confirmed by tryptase immunohistochemistry (or other special stains).
Minor criteria
a. In biopsy sections of bone marrow or other extracutaneous organs, more than 25% of the mast cells in the infiltrate are spindle-shaped or have atypical morphology, or, of all mast cells in bone marrow aspirates smears, more than 25% are immature or atypical mast cells.
b. Detection of KIT mutation at codon 816 in bone marrow, blood or other extracutaneous organ(s).
c. Mast cells in bone marrow, blood or other extracutaneous organs that co-express CD117 with CD2 and/or CD25.
d. Serum total tryptase persistently >20 ng/ml (if there is an associated clonal myeloid disorder, this parameter is not valid).
The diagnosis of SM may be made if one major and one minor criterion are present, or, if three minor criteria are met.

[1] Adapted from 6.

tissues, such as the skin, gastrointestinal tract, or bone marrow, are not diagnostic because such findings are also found in normal inflammatory or reactive process. Likewise, while plasma or urinary levels of histamine are frequently increased in systemic mastocytosis (48), the solitary finding of increased levels of histamine or histamine metabolites may reflect any of a number of other situations, such as anaphylaxis or response to unusual immunologic stimuli. Similarly, serum tryptase may be elevated after anaphylaxis, and may sometimes be normal in the presence of marrow disease. Thus, no single laboratory test can establish the diagnosis of mastocytosis. Rather, the demonstration of mast cell mediators in blood or urine simply prompts the clinician to investigate further for the presence of abnormal collections of mast cells.

Therapy

No mast cell ablative therapy has been yet reported for mastocytosis and there is no evidence that symptomatic therapy substantially alters the course of disease. A practical concern in therapy for both cutaneous and systemic mast cell disease is the avoidance of triggering factors such as temperature extremes, physical exertion, or in some cases, the ingestion of agents such as ethanol, nonsteroidal anti-inflammatory drugs or opiate analgesics. Physical trauma to lesions or environmental factors may also trigger acute episodes, with reports of hypotension after *Hymenoptera* stings and exposure to iodinated contrast materials (49). Epinephrine remains the drug of choice in the treatment of hypotension, either idiopathic or induced by environmental factors. Patients with mast cell disease and a history of hypotensive episodes should be advised to carry epinephrine-filled syringes and taught to self-medicate. These patients may also benefit from the concurrent use of H_1 and H_2 antihistamines prophylactically. For a review of therapeutic approaches applied to mastocytosis, see reference 50.

Classic as well as newer non-sedative H_1 histamines are used to decrease irritability of the skin and mitigate symptoms of pruritis. Amelioration can be achieved through the use of antihistamines but rarely does total ablation of signs and symptoms occur. Hydroxyzine and diphenhydramine are two potent H_1 antihistamines found to be quite useful. Frequently the dose-limiting side effect of antihistamine therapy is sedation. Patients sensitive to the sedative effects of antihistamines may benefit from the use of newer nonsedative antihistamines. For patients who continue to have significant disease symptoms while on H_1 antihistamines, the combination of H_1 and H_2 antagonists has been shown to be at times effective in relieving pruritis and wheal

formation. H_2 histamines such as ranitidine and famotidine and proton pump inhibitors (omeprazole) have been useful in the treatment of gastritis and peptic ulcer disease associated with mastocytosis.

Oral administration of disodium cromoglycolate has been reported to reduce pruritis and wheal formation in UP in patients with or without systemic disease (51). This agent has also been reported to be of benefit in cutaneous mast call disease in children and infants and proved most useful in some patients for the control of gastrointestinal complaints (52). Sometimes other symptoms such as headache and bone pain have also been reported to improve with the use of cromolyn sodium. Diphosphonates when appropriate are used in the treatment of mastocytosis associated ostopenia (53).

While systemic corticosteroids have not been shown to be effective in the treatment of cutaneous mastocytosis, topical administration or intralesional injections of corticosteroids have resulted in symptomatic and cosmetic improvement. Caution must be exercised with repeated or extensive application of corticosteroids, as this may result in cutaneous atrophy or adrenocortical suppression (54). The oral administration of 8-methoxypsoralen plus ultraviolet A (PUVA) photochemotherapy has resulted in a decrease in pruritis and wheal formation in patients with UP with or without systemic disease (55). Relapses occurring 3–6 months after cessation of therapy are common. Patients sometimes report a decrease in cutaneous lesions after exposure to natural sunlight. There are no controlled studies using ultraviolet B photography.

Systemic corticosteroids have been useful in decreasing the malabsorption and ascites in when present. In adults, oral prednisone (40–60 mg/day) usually results in a decrease in symptoms over a 2–3 week period. After initial improvement, steroids can frequently be tapered to an alternate-day dosing regimen. However, with time, the ascites frequently recurs. It has been suggested that selected patients may benefit from a portacaval shunt (56).

Aspirin and other non-steroidal anti-inflammatory agents have been useful in some patients whose primary manifestation is recurrent episodes of flushing or syncope, or both. The use of these agents may be problematic in patients with significant ulcer disease. Some patients may worsen with aspirin.

A small percentage of patients with systemic mast cell disease may have a syndrome mimicking a non-Hodgkin lymphoma, an aggressive myeloproliferative disease, or rarely an overt nonlymphocytic leukemia (30,57). Occasional patients have been reported with systemic mast cell disease associated with primary mediastinal germ cell tumor

(58). In this group of patients, traditional chemotherapy directed toward their neoplastic process may be appropriate. Radiotherapy has been used in limited patients to control local disease (59).

Splenectomy has been performed on a number of patients with severe aggressive forms of mastocytosis, in an attempt to improve their limiting cytopenias (60). With splenectomy, survival increased by an average of 12 months. Patients who had undergone splenectomy appeared better able to tolerate chemotherapy.

Prognosis

Patients who present with cutaneous disease, flushing disorders, or limited extracutaneous organ involvement frequently have an indolent course requiring chronic medical management. Few if any of these cases have been documented to progress into a more advanced form of the disease (30). In contrast to the clinical course of patients with limited disease, a prospective analysis of 46 patients identified elevated lactate dehydrogenase levels, late age of onset, and the presence of a significant hematologic abnormality (such as myeloproliferative, myelodysplastic, or overt leukemic picture) as indicators of a poor prognosis and shortened survival (30). Of the parameters studied by multivariate analysis, only late age at onset of symptoms and elevated serum lactate dehydrogenase levels were found to be predictive of a poor prognosis. Other groups have also identified the presence of a myeloproliferative or myelodysplastic blood picture as conferring a poorer prognosis.

Summary and Future Directions

In addition to the therapeutic modalities already mentioned, some innovative therapies on the horizon are worth discussing separately. Mastocytosis involves both mast cell hyperplasia and systemic mediator release. An approach using an "antiproliferative" mediator to treat mastocytosis is attractive and may warrant investigation. Clinical responses to IFN-α2B have been reported and have been mixed (61,62). Cladribine (2-chlorodeoxyadenosine), a purine nucleoside analogue, has been used to induce clinical remissions in some patients with more aggressive forms of mastocytosis. Cladribine may be considered for treating those with aggressive forms of mastocytosis who have interferon-α resistant advanced disease (63). In other patients, bone marrow transplantation may have the potential of allowing long-term engraftment of healthy bone marrow progenitors along with provision for a graft vs. leukemia effect. There is limited data applying

this approach to patients with aggressive forms of mastocytosis (64,65).

Therapeutic options for patients with mastocytosis with an associated clonal hematologic non-mast cell lineage disease (SM-AHNMD), aggressive systemic mastocytosis (ASM) and mast cell leukemia (MCL) are directed to treating the associated hematologic disorder. Various chemotherapeutic regimens have been tried with mixed success and are summarized elsewhere (Review #50). Regardless of the regimens employed, relatively short partial remissions were noted in the majority of patients treated.

Tyrosine kinase inhibitors are under investigation as a potential therapeutic class to interfere with mast cell proliferation and survival. It has been suggested that the codon 816 Kit mutation is suppressed by imatinib (STI571). However, additional studies on effects of these tyrosine kinase inhibitors, including STI571 on activation of the catalytic domain of Kit indicated that wild type Kit was more effectively inhibited than Kit with a codon 816 mutation (66,67). These data indicate that the catalytic domain of Kit with a codon 816 mutation is characterized by an activation mechanism resistant to tyrosine kinase inhibitors. It is not unreasonable to postulate that the patient population with mastocytosis that is most likely to respond to these tyrosine kinase inhibitors will comprise those that lack a codon 816 activating mutation. Protein kinase inhibitors targeted to mutated Kit are under development.

REFERENCES

1. Nettleship E, Tay W: Rare forms of urticaria. Br Med J 1869;2:323.
2. Ehrlich P: Bëitrage Zur theoretic and Praxis de histologischer Färbung (Ph.D thesis, Leipzig, Germany: University of Leipzig; 1878).
3. Unna PG: Bëitrage Zur Anatomic und Pathogenese der Urticaria Simplex und Pigmentosa. Mschr Prakt Dermatol, Suppl Dermatol Stud 1887;3:9.
4. Sangster A: An anomalous mottled rash, accompanied by pruritis, factious urticaria and pigmentation, "urticaria pigmentosa (?)." Trans Clin Soc London 1878;11:161.
5. Ellis JM: Urticaria Pigmentosa: a report of a case with autopsy. Arch Pathol 1949;48:426.
6. Valent P, Horny H, Longley BJ, et al: Mastocytosis. In: World Health Organization Classification of Tumors: Pathology and Genetics in Tumours of Haematopoietic and Lymphoid Tissues. Jaffe ES, Harris NL, Stein H, Varderman JE, eds: IARC Press, Lyon, 2001;291–302.
7. Kitamura Y, Go S, Hatanaka S: Decrease of mast cells in W/W^V mice and their increase by bone marrow transplantation. Blood 1978;52:447.
8. Geissler EN, Ryan MA, Housman DE: The dominant-white spotting (W) locus of the mouse encodes the c-kit protooncogene. Cell 1988;55:185.
9. Kirshenbaum AS, Goff JP, Kessler SW, et al: Effect of IL-3 and stem cell factor on the appearance of human basophils and mast cells from CD34+ pluripotent progenitor

cells. J Immunol 1992;148:772–777.
10. Anderson DM, Lyman SD, Baird A, Wignall JM, Eisenman J, Rauch C, March CJ, Boswell HS, Gimpel SD, Cosman D, Williams DE: Molecular cloning of mast cell growth factor, a hematopoietin that is active in both membrane bpund and soluble forms. Cell 1990;63:235.
11. Metcalfe DD, Baram D, Mekori YA: Mast Cells. Physiol Rev 1997;77:1033.
12. Irani AA, Schechter NM, Craig SS, et al: Two types of human mast cells that have distinct neutral protease compostions. PNAS 1986;83:4464.
13. Rottem M, Okada T, Goff JP, Metcalfe DD: Mast cells cultured from the peripheral blood of normal donors and patients with mastocytosis originate from a CD34+/Fc epsilon RI-cell population. Blood 1994;84:2489.
14. Nagata H, Worobec AS, Oh CK, et al: Identification of point mutation in the catalytic domain of the proto-oncogene c-kit in the peripheral blood mononuclear cells of patients with mastocytosis. PNAS 1995;92:10560.
15. Longley BJ, Metcalfe DD, Tharp M, et al: Activating and dominant inactivating c-kit catalytic domain mutations in distinct clinical forms of human mastocytosis. PNAS 1999;96:1609.
16. Akin C, Kirshenbaum AS, Semere T, et al: Analysis of the surface expression of c-kit and occurrence of the c-kit Asp816Val activating mutation in T cells, B cells, and myelomonocytic cells in patients with mastocytosis. Exp Hematol 2000;28:140.
17. Pignon JM, Giraudier S, Duquesnoy P, Jouault H, Imbert M, Vainchenker W, Vernant JP, Tulliez M. A new c-kit mutation in a case of aggressive mast cell disease. Br J Haematol 1997;96:374–376.
18. Akin C, Fumo G, Yavuz AS, Lipsky PE, Neckers L, Metcalfe DD: A novel form of mastocytosis associated with a transmembrane c-Kit mutation and response to imatinib. Blood 2003;103:3222–3225.
19. Pullarkat VA, Pullarkat ST, Calverley DC, Brynes RK. Mast cell disease associated with acute myeloid leukemia: detection of a new c-kit mutation Asp816His. Am J Hematol 2000;65:307–309.
20. Valent P, Akin C, Sperr WR, et al: Mastocytosis: pathology, genetics, and current options for therapy. Leuk Lymphoma 2004;45:in press.
21. Pardanani A, Ketterling RP, Brockman SR, Flynn HC, Paternoster SF, Shearer BM, Reeder TL, Li CY, Cross NC, Cools J, Gilliland DG, Dewald GW, Tefferi A, CHIC2 deletion, a surrogate for FIP1L1-PDGFRA fusion, occurs in systemic mastocytosis associated with eosinophilia and predicts response to imatinib mesylate therapy. Blood 2003;102:3093–3096.
22. Swolin B, Rodjer S, Roupe G: Cytogenetic studies and in vitro colony growth in patients with mastocytosis. Blood 1987;70:1928–1932.
23. Lishner M, Confino-Cohen R, Mekori YA, Feigin M, Manor Y, Goldberg A, Ravid M, Amiel A. Trisomies 9 and 8 detected by fluorescence in situ hybridization in patients with systemic mastocytosis. J Allergy Clin Immunol 1996;98:199–204.
24. Daley T, Metcalfe DD, Akin C: Association of the Q576R polymorphism in the interleukin-4 receptor alpha chain with indolent mastocytosis limited to the skin. Blood 2001;98:880–882.
25. Escribano L, Orfao A, Diaz-Agustin B, Villarrubia J, Cervero C, Lopez A, et al: Indolent systemic mast cell disease in adults: immunophenotypic characterization of bone marrow mast cells and its diagnostic implication. Blood 1998;91:2731–2736.
26. Webb TA, Li C-Y, Yam LT: Systemic mast cell disease: a clinical and hematopathologic study of 26 cases. Cancer 1982;49:927.
27. Horny H-P, Parwaresch MR, Lennart K: Bone marrow findings in systemic mastocytosis. Hum Pathol 1985;16:808.

28. Kettelhut BV, Parker RI, Travis WD, Metcalfe DD: Hematopathology of the bone marrow in pediatric cutaneous mastocytosis: A study of 17 patients. Am J Clin Pathol 1989;91:558.
29. Brockow K, Scott LM, Worobec AS, et al: Regression of urticaria pigmentosa in adult patients with systemic mastocytosis: Correlation with clinical patterns of disease. Arch Dermatol 2002;138:785.
30. Lawrence JB, Friedman GB, Travis WD, et al: Hematologic manifestations of systemic mast cell disease: a prospective study of laboratory and morphologic features and their relation to prognosis. Am J Med 1991;91:612.
31. Horny H-P, Kaiserling E, Parwaresch MR, Lennert K: Lymph node findings in generalized mastocytosis. Histiopath 1992;21:439.
32. Travis WD, Li C-Y: Pathology of the lymph node and spleen in systemic mast cell disease. Modern Pathology 1988;1:4.
33. Mican JM, DiBisceglie AM, Fong T-L, et al: Hepatic involvement in mastocytosis: clinicopathologic correlations in 41 cases. Hepatology 1995;22:1163.
34. Travis WD, Li C-Y, Hoagland HC, et al: Mast cell leukemia: report of a case and review of the literature. Mayo Clin Proc 1986;61:957.
35. Rogers MP, Bloomingdale K, Murawski BJ, et al: Mixed organic brain syndrome as a manifestation of systemic mastocytosis. Psychosomatic Med 1986;48:437.
36. Caplan RM: The natural course of urticaria pigmentosa: analysis and follow-up of 112 cases. Arch Dermatol 1963;87:146.
37. Brockow K, Akin C, Huber M, Metcalfe DD: Assessment of the extent of cutaneous involvement in children and adults with mastocytosis: relationship to symptomatology, tryptase levels, and bone marrow pathology. J Amer Acad Dermatol 2003;48:508.
38. Jelinak JE: Urticaria pigmentosa bei drei leibichen Brüdern. Hautazart 1970;21:303.
39. Bazex A, Duprét A, Christol B, Andrieu H: Les mastocytosis familiales: presentation de deux observations: revue générale: intérêt nosologique. Ann Dermatol Syphilol 1971;98:241.
40. Von Weber K: Urticaria pigmentosa bei eineiigen Zwillingen. Dermatol Monatssschr 1973;159:258.
41. Rockoff AS: Urticaria pigmentosa in identical twins. Arch Dermatol 1978;114:1227.
42. Soter NA: Mastocytosis of the skin. Hem Onc Clinics of North America 2000;14:537.
43. Klaus SN, Winkelmann RK: Course of urticaria pigmentosa in children. Arch Dermatol 1962;86:68.
44. Cherner JA, Jensen RT, Dubois A, et al: Gastrointestinal dysfunction in systemic mastocytosis. Gastroenterology 1988;95:657.
45. Fallon MD, Whyte MP, Teitelbaum SL: Systemic mastocytosis associated with generalized osteopenia. Hum Pathol 1981;12:813.
46. Chen CC, Andrich MP, Metcalfe DD. A retrospective analysis of bone scans abnormalities in mastocytosis: correlation with disease category and prognosis. J Nuclear Med 1994;35:1471.
47. Travis WD, Li C-Y, Bergstralh EF, et al: Systemic mast cell disease: Analysis of 58 cases and literature review. Medicine 1988;67:345.
48. Friedman BS, Steinberg S, Meggs WJ, et al: Analysis of plasma histamine levels in patients with mast cell disorders. Am J Med 1989;87:649.
49. Müller UR, Horat W, Wuthrsch B, Conroy M, Reisman RE: Anaphylaxis after hymenoptra stings in three patients with urticaria pigmentosa. J Allergy Clin Immunol 1983;72:685.
50. Worobec AS: Treatment of systemic mast cell disorders. Hem Onc Clinics of North

America 2000;14:659.
51. Soter NA, Austen KF, Wasserman ST: Oral disodium cromoglycolate in the treatment of systemic mastocytosis. NEJM 1979;310:465.
52. Welch EA, Alper JC, Bogaars H, Farrell DS: Treatment of bullous mastocytosis with disodium cromoglycolate. J Am Acad Dermatol 1983;9:349.
53. Cundy T, Beneton MNC, Darby AJ, et al: Osteopenia in systemic mastocytosis: natural history and responses to treatment with inhibitors of bone resorbtion. Bone 1987;8:149.
54. Barton J, Lauker RM, Schecter NM, Lazarus GS: Treatment of urticaria pigmentosa with corticosteroids. Arch Dermatol 1985;121:1516.
55. Vella Briffa D, Eady RAJ, James MP, et al: Photochemotherapy (PUVA) in the treatment of urticaria pigmentosa. Br J Dermatol 1983;109:67.
56. Bonnet P, Smadja C, Szekely A-M, et al: Intractable ascites in systemic mastocytosis treated with portal diversion. Dig Dis Sci 1987;32:209.
57. Hutchinson RM: Mastocytosis and co-existent Non-Hodgkin's Lymphoma and myeloproliferative disorders. Leuk Lymphoma 1992;7:29.
58. Chariot P, Monnet I, LeLong F, et al: Systemic mast cell disease associated with primary mediastinal germ cell tumor. Am J Med 1991;90:381.
59. Johnstone PA, Mican JM, Metcalfe DD, DeLaney TF: Radiotherapy of refractory bone pain due to systemic mast cell disease. Am J Clin Oncology 1994;17:328.
60. Friedman B, Darling G, Norton J, et al: Splenectomy in the management of systemic mast cell disease. Surgery 1990;107:94.
61. Kluin-Nelemans HC, Jansen JH, Breukelman H, et al: Response to interferon Alfa-2b in a patient with systemic mastocytosis. NEJM 1992;329:619.
62. Worobec AS, Kirshenbaum AS, Schwartz LB: Treatment of three patients with mastocytosis with interferon alpha-2b. Leukemia and Lymphoma 1996;22:501.
63. Tefferi A, Chin-Yang L, Butterfield JH, Hoagland HC. Treatment of systemic mast cell disease with cladribine. New Engl J Med 2000;344:307.
64. Prezpiorka D, Giralt S, Khouri I, et al: Allogeneic marrow transplantation for myeloproliferative disorders other than chronic myelogenous leukemia: review of forty cases. Am J Hematol 1998;57:23.
65. Ronnov-Jessen D, Lovgreen Nielson P, Horn T: Persistence of systemic mastocytosis after allogeneic bone marrow transplantation in spite of complete remission of the associated myelodysplastic syndrome. Bone Marrow Transplant 1991;8:413.
66. Ma Y, Zeng S, Metcalfe DD, et al: The c-kit mutation causing human mastocytosis is resistant to STI571 and other kit kinase inhibitors; kinases with enzymatic site mutations show different inhibitor sensitivity profiles than wild-type kinases and those with regulatory-type mutations. Blood 2002;99:1741.
67. Akin C, Brockow K, D'Ambrosio D, et al: Tyrosine kinase inhibitor STI571 does not show preferential cytotoxicity on neoplastic human mast cells bearing codon 816 c-kit mutations. Exp Hematol, in press.

DISCUSSION

MacKowiak, Baltimore: Could you give us some follow-up on that 25 year old woman's treatment?

Metcalfe, Bethesda: I can. We continue to treat her on a special patient protocol. Recently, she had to be taken off imatinib for surgery. For a while her serum tryptase levels stayed down, then started to climb. Following surgery we re-instituted imatinib

and again observed a decrease in serum tryptase. There are other tyrosine kinase inhibitors in trials now that offer promise of inhibiting Kit with a codon 816 mutation. So we are hoping that there's going to be additional therapeutic options for this young woman with time. A rare patient with a malignant clone that responds to imatinib will see that malignant clone apparently eliminated with imatinib, but most of the time we believe its use buys time.

Mackkowiak: How's she feeling?

Metcalfe: She feels great. She came in, as you can see by her emails, felling horrible. Within days she began to feel stronger, and with time her peripheral blood picture improved. No tumor lysis syndrome was observed. It looks like when you induce mast cell apoptosis, there is phagocytosis of these dying mast cells by surrounding cells, which in turn limits mediator release and constitutional symptoms. We do institute therapy under close observation, concerned about possible tumor lysis with hypotension, but we have not observed such an outcome.

Clarkson, New York: You already mentioned that you're aware there are a number of c-kit and c-abl tyrosine kinase inhibitors. We've been working with some of the pyrido-pyrimidines and other inhibitors, and there is quite a different spectrum of resistance to point mutations in the ATP binding pocket of abl exhibited by different inhibitors. The most potent pyrido-pyrimidines are ~80–100 fold more inhibitory to abl and bcr-abl than Gleevec and are also more inhibitory to c-kit, pdgfr, and several src family members. Minor changes in the structure of the inhibitors can result in significant changes in the sensitivity of the different kinases.

Metcalfe: You are absolutely correct. We also believe we can obtain some idea of their efficacy against specific mutations in Kit by testing a given agent's effect on the survival of bone-marrow mast cells in culture. We gate on mast cells in these ex-vivo cultures and are able to see if a given agent causes mast cell apoptosis in diseased mast cells vs. mast cells from normal donors. We believe this is a good example of pharmacogenomics.

Stevenson, Stanford: Very nice presentation, Dean. In newborn medicine we very rarely see mastocytosis. Have you had a chance to look at any of the younger individuals to see if they are biologically unique, compared to the older children? The following question is something you mentioned very briefly. Could you give me some brief insight into the trafficking protein?

Metcalfe: About half of the cases of mastocytosis present in childhood. Most cases are diagnosed before the age of six months. The later the presentation the higher the associated prevalence of associated hematologic disorders, so early presentation is better. Most children have cutaneous disease, but some have an adult pattern of disease and where a codon 816 activating mutation may be present. Some children with only cutaneous disease "outgrow" the disease, so whether or not this is some kind of overproduction of stem cell factor or something else, we do not know.

Adkinson, Baltimore: Dean, the genetic mutations that you described clearly explain the proliferation of large numbers of mass cells in mastocytosis. Do you have any understanding about how these mutations could also make the cells more fragile and subject to the spontaneous release of mediators that is apparently responsible for the anaphylactoid reactions seen clinically?

Metcalfe: It may be that codon 816 activating mutations also allow mast cells to degranulate to specific and non-specific stimuli with a lower threshold. That is, these mast cells are "twitchy." We are working on this hypothesis now with in-vitro modeling.